

Claims

1. A method of identifying a cell colony which expresses a soluble variant of a target protein, which method comprises:
  - (a) subjecting said cell colony to conditions which are capable of causing lysis thereof;
  - (b) filtering the lysate of step (a) through a filter having pores which allow only soluble proteins to pass through the filter; and
  - (c) detecting target protein which has passed through the filter, wherein the target is not detected on the basis of its own enzymatic activity.
2. The method of claim 1 wherein the lysis of step (a) is native lysis.
3. The method of claim 2 wherein native lysis is carried out by freeze thawing colonies.
4. The method of claim 2 or 3 wherein native lysis is carried out using native lysis buffer.
5. The method of any one of claims 1 to 4 wherein the target protein is fused to a protein or polypeptide tag.
6. The method of any one of claims 1 to 5, wherein soluble proteins in the filtrate are identified using antibodies and/or fusion tags.
7. The method of claim 5 or 6 wherein the fusion tag is His.
8. The method of any one of claims 5 to 7 wherein the tag acts as the substrate in an enzymatic detection method for detecting the target protein in step (c) of claim 1.

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9. The method of any one of claims 1 to 8 wherein step (c) is a non-enzymatic detection method.
10. The method of any one of claims 1 to 9 wherein said filter has a pore size between 0.1 and 1.5  $\mu\text{m}$ .
11. The method of any one of claims 1 to 10 wherein said colonies are lifted from their growth media on the filter used in step (b).
12. The method of claim 11, wherein said colonies are lifted prior to the lysis of step (a).
13. The method of any one of claims 1 to 12, wherein filtration step (b) includes the application of a force to the filter carrying the colonies.
14. The method of any one of claims 1 to 13, wherein proteins in the filtrate from filtration step (b) are captured on a solid support prior to detection step (c).
15. The method of any one of claims 1 to 14 wherein said protein is a membrane protein.
16. A method of identifying a cell colony expressing a soluble variant of a membrane protein, which method comprises:
  - (a) subjecting one or more colonies of cells to conditions which are capable of causing lysis thereof;
  - (b) filtering the lysate of step (a) through a filter having pores which allow only soluble proteins to pass through the filter, thereby generating a filtrate containing soluble membrane proteins.
17. A kit for use in the methods of any one of claims 1 to 15 comprising:
  - (a) a filter having pore sizes which only allow

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soluble proteins to pass through the filter;

(b) said support; and optionally

(c) reagents for use in native lysis of the cell colonies.